

## AMENDMENT TO THE SPECIFICATION

**On page 2, line 8 of the specification, please replace the background of the invention with following amended paragraph:**

This application is a ~~continuation~~ divisional of ~~patent application~~, U.S. Patent Application Serial No. 09/662,270, filed on September 14, 2000, now abandoned, which is a divisional ~~application~~ of U.S. Patent Application Serial No. 09/097,199, filed on June 12, 1998 and issued as U.S. Patent No. 6,218,529 which was a continuation-in-part of U.S. Patent Application Serial No. 08/692,787 filed July 31, 1996 and issued as U.S. Pat. Patent No. 5,882,864, which claims the benefit of U.S. provisional applications 60/001,655, filed ~~Jul.~~ July 31, 1995, and 60/013,611, filed Jan. 11, 1996, both now abandoned. ~~The entire text of the above-referenced disclosure is specifically incorporated by reference herein without disclaimer.~~ The entire text of U.S. Patent Application Serial No. 09/662,270, U.S. Patent Application Serial No. 09/097,199 (now U.S. Patent No. 6,218,529) and U.S. Patent Application Serial No. 08/692,787 (now U.S. Patent No. 5,882,864) is incorporated herein by reference.

**Please replace the paragraph beginning on line 1 of page 27, with the following amended paragraph:**

In certain broad applications of the invention, the gene sequence encoding the polypeptide is analyzed to detect putative transmembrane sequences. Such sequences are typically very hydrophobic and are readily detected by the use of standard sequence analysis software, such as MacVector<sup>TM</sup> (IBI, New Haven, CT). The presence of transmembrane sequences is often deleterious when a recombinant protein is synthesized in many expression systems, especially E. coli, as it leads to the production of insoluble aggregates which are difficult to renature into the native conformation of the protein. Deletion of transmembrane sequences typically does not significantly alter the conformation of the remaining protein structure.

**Please replace the paragraph beginning on line 13 of page 27, with the following amended paragraph:**

Computer sequence analysis may be used to determine the location of the predicted major antigenic determinant epitopes of the polypeptide. Software capable of carrying out this analysis is readily available commercially, for example MacVector<sup>TM</sup> (IBI, New Haven, Conn.). The software typically uses standard algorithms such as the Kyte/Doolittle or Hopp/Woods methods for locating hydrophilic sequences may be found on the surface of proteins and are, therefore, likely to act as antigenic determinants.

**Please replace the paragraph beginning on line 5 of page 51, with the following amended paragraph:**

"Under conditions effective to allow immune complex (antigen/antibody) formation" means that the conditions preferably include diluting the antigens and antibodies with solutions such as BSA, bovine gamma globulin (BGG) and phosphate buffered saline (PBS)/Tween<sup>TM</sup>. These added agents also tend to assist in the reduction of nonspecific background.

**Please replace the paragraph beginning on line 14 of page 51, with the following amended paragraph:**

Following all incubation steps in an ELISA, the contacted surface is washed so as to remove non-complexed material. A preferred washing procedure includes washing with a solution such as PBS/Tween<sup>TM</sup>, or borate buffer. Following the formation of specific immune complexes between the test sample and the originally bound material, and subsequent washing, the occurrence of even minute amounts of immune complexes may be determined.

**Please replace the paragraph beginning on line 4 of page 85, with the following amended paragraph:**

The cell lines were propagated in RPMI-1640 (GIBCO-BRL, Inc.) supplemented with 10% fetal bovine serum, 5 units/ml penicillin G, 5 pg/ml streptomycin, and Fungizone<sup>TM</sup> according to the supplier's directions. All antibiotics were purchased from GIBCO-BRL, Inc. Cells were harvested in late log phase of growth. RNA was isolated by the guanidinium thiocyanate method (Chomczynski and Sacchi, 1987). RNA was also isolated from solid prostate tumors by guanidinium thiocyanate extraction (Chomczynski and Sacchi, 1987), after the tumors were frozen and ground to a powder in liquid nitrogen.

**Please replace the paragraph beginning on line 22 of page 88, with the following amended paragraph:**

Cycling parameters: 30 cycles of 94°C for 1 min; 55°C for 1 min; and 72°C for two min. Thermocyclers were either the MJ research thermocycler or the Stratagene Robocycler<sup>TM</sup>.

**Please replace the paragraph beginning on line 19 of page 103, with the following amended paragraph:**

The final major barrier to quantifying relative mRNA abundances with RT-PCR is tube to tube variability in PCR. This can result from many factors, including unequal heating and cooling in the thermocycler, imperfections in the PCR tubes and operator error. To control for this source of variation, the Cole-Parmer<sup>TM</sup> digital thermocouple Model # 8402-00 was used to calibrate the thermocyclers used in these studies. Only slight variations in temperature were observed. To rigorously demonstrate that PCR tube to tube variability was not a factor in the studies described above, 24 duplicate PCRs for  $\beta$ -actin using the same cDNA as template were performed. These PCR tubes were scattered over the surface of a 96 well thermocycler, including the corners of the block where it might be suspected the temperature might deviate from other areas. Tubes were collected at various cycle numbers. Nine tubes were collected at 21 cycles. Nine tubes were collected at 24 cycles, and six tubes were collected at 27 cycles. Quantitation of the intensities of the resulting bands with the IS 1000 system determined that the standard error of the mean of the

PCR product abundances was  $\pm 13\%$ . This is an acceptably small number to be discounted as a major source of variability in an RT-PCR assay.

**Please replace the paragraph beginning on line 8 of page 119, with the following amended paragraph:**

American Cancer Society-Facts and Figures-1998, which may be found on the world wide web at [www.cancer.org/statistics/98cff/98prosta.html](http://www.cancer.org/statistics/98cff/98prosta.html).